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Protective roles of inorganic nitrate in health and diseases

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Abstract

Various beneficial biological activities of inorganic nitrate have been revealed in recent decades. Oral bacteria can reduce nitrate to nitrite, which is further reduced to nitric oxide (NO) in the body; this process is known as the nitrate-nitrite-NO pathway. Sialin is a mammalian membrane nitrate transporter that transports nitrate to the salivary glands and secretes it into the oral cavity through the saliva. Recent studies have indicated that nitrate has a protective effect on the salivary glands and other organs by regulating the expression of sialin and maintaining microbial homeostasis. Through the nitrate-nitrite-NO pathway, nitrate can act as a reservoir of NO in vivo and perform a variety of NO-like bioactivities, such as promoting exercise performance, protecting the digestive system, lowering blood pressure, and assisting in tumor treatment. This paper reviews the sources, functions, and possible mechanisms of inorganic nitrate, and discusses the protective role that nitrate promises to play in health and diseases.

Keywords: Inorganic nitrate, Nitrite, Nitric oxide, Sialin, Microbial homeostasis, Nitrate-nitrite-NO pathway

1 Introduction

Nitrate is an inorganic anion widely distributed in nature (Gassara et al., 2016). Historically, nitrate was considered an inert, metabolic end product of nitric oxide (NO). However, in recent decades, the biological activity of nitrate as a natural dietary nutrient has been widely studied. The application of nitrate as a medicine can be traced back to ancient China in the eighth century AD, when a manuscript from Mogao Grottoes documented the use of nitrate in the treatment of acute cardiovascular diseases (Lundberg et al., 2008). In the early twentieth century, bismuth nitrate was utilized for the treatment of peptic ulcer disease and diarrhea. Interestingly, doctors at the time found that high doses of nitrate were associated with a side effect of lowering blood pressure, suggesting that inorganic nitrates might play a role in treating hypertension (Gorbach, 1990). Later, it was discovered that nitrate could be reduced by bacteria, and the nitrate-nitrite-NO pathway was proposed (Kapil et al., 2020). With further research, various other functions of nitrate have been discovered. This review summarizes the protective roles of inorganic nitrate in health and disease.

2 Nitrate sources and metabolism

Nitrate is widely distributed in the natural environment, including in food, water, air, and soil (Lundberg et al., 2008). Nitrate exists abundantly in different organs of the human body. The nitrate level in the plasma is usually in the range of about 20 to 40 μmol/L. The mean concentrations of nitrate in saliva and urine of healthy individuals are on the scale of millimole per liter. High concentrations of nitrate are also present in organs such as the heart, liver, and muscle (Lundberg & Govoni, 2004; Xia et al., 2003). There are two sources of nitrate in the human body: endogenous and exogenous sources.

The endogenous source of nitrate is the oxidation of NO. The human body uses the classical NO synthesis pathway, where nitric oxide synthase (NOS) acts on L-arginine and molecular oxygen to synthesize NO (Lundberg et al., 2008). There are three isoforms of NOS: inducible NOS (iNOS or NOS-2), neuronal NOS (nNOS

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or NOS-1), and endothelial NOS (eNOS or NOS-3), each of which affects different locations and conditions. nNOS and eNOS are collectively called constitutive NOS (cNOS), which exists persistently in a healthy body. In response to physical or receptor stimulation, cNOS produces a limited quantity of NO (Iwata et al., 2020). In healthy people, daily synthesis of endogenous nitrate of up to 0.5 to 1 mmol occurs, which amounts to 25% of the total nitrate in the human body (Green et al., 1981). In contrast, in patients with systemic inflammatory disorders such as sepsis and severe gastroenteritis, iNOS is highly induced, and NO levels are remarkably increased (Crawford et al., 2004). NO can be spontaneously or catalytically oxidized to nitrite by the multi-copper oxidase and NO oxidase ceruloplasmin in the plasma (Shiva et al., 2006). Furthermore, NO and nitrite can rapidly react with oxyhemoglobin to produce nitrate and methemoglobin in the blood (Moncada & Higgs, 1993; Weitzberg & Lundberg, 2013). NO has a short half-life in the order of seconds. Nitrite also has a rather short half-life; with a half-life of 110 s, nitrite is quickly converted to nitrate in the whole blood, whereas it is relatively stable in the plasma with a half-life of 20–30 min (Ma et al., 2018). Plasma nitrate has a long half-life of 6 h. Circulating plasma nitrate levels remained significantly high 24 h after a single dose (Kapil et al., 2010).

The exogenous source of nitrate is mainly the diet, especially vegetables. According to a database on the nitrate content of vegetables, the median concentrations of nitrate vary from 12 mg/kg (corn) to 4240 mg/kg (Chinese flat cabbage) (Blekkenhorst et al., 2017). Green leafy vegetables, such as spinach, lettuce, broccoli, and beetroot, provide the most important supply of exogenous nitrate, accounting for more than 80% of it. The World Health Organization (WHO) recommends that a person should consume 400 g of mixed vegetables (containing about 2.5 mmol nitrate) per day (Kapil et al., 2020). Some diets, such as the Mediterranean diet and traditional Asian diet, provide more nitrate than other diets (Sobko et al., 2010). In addition, drinking water and canned meat also contain nitrate, accounting for less than 20% of daily dietary intake (Hord et al., 2009). Compared with endogenous sources, diet is the main source of nitrate for people.

After a nitrate-rich diet, nitrate is rapidly absorbed in the digestive tract and enters the systemic circulation. Nitrate has almost 100% bioavailability because it does not transform upon first-pass metabolism (van Velzen et al., 2008). An increase in plasma nitrate can be detected within 15 min and reach a peak at around 30 to 60 min (McKnight et al., 1997). Following ingestion by the digestive tract, approximately 25% of the nitrate in the plasma is actively absorbed by the salivary glands through sialin, a nitrate transporter. In addition, approximately two-thirds of the nitrate is excreted in the urine, and a small amount is excreted in the sweat and feces. Elevated nitrate levels can be detected in the saliva within 20 to 60 min of nitrate supplementation. Salivary nitrate concentrations can reach 10–20 times higher than plasma concentrations (Qin et al., 2012). The reuptake process is called the enterosalivary circulation of nitrate (Duncan et al., 1995). In this process, the parotid glands are the most important organs. Sialin, a transmembrane protein, also plays a significant role. Sialin is highly expressed in the parotid gland, brain, liver, pancreas, kidney, and cardiovascular system, and it is most abundantly expressed in the acinar cells of the parotid gland (Qin et al., 2012). Nitrate transport into salivary gland cells through sialin is the key step for the nitrate-nitrite-NO pathway and maintenance of NO homeostasis in the human body, which is important for health and disease prevention (Lundberg, 2012) (Fig. 1).

High concentrations of salivary nitrate are partially reduced to nitrite and NO by oral bacteria. After swallowing, nitrate and nitrite are reabsorbed into the blood via the intestinal mucosa, and nitrite is either quickly oxidized to nitrate in the peripheral blood or reduced to NO (Jones et al., 2018; Lundberg et al., 2018). This process is known as the nitrate-nitrite-NO pathway. Through the nitrate-nitrite-NO pathway, nitrate is thought to serve as a reservoir of NO in the body, performing an NO-like function in a variety of physiological and pathological conditions (Lundberg et al., 2008).

Nitrate reductase is deficient in humans and other mammals. Xanthine oxidoreductase (XOR) is the only known nitrate reductase in mammals. However, with a nitrate-rich diet, the normal level of nitrite in the plasma can be increased 4–5-fold (Ma et al., 2018). Nitrite is present in saliva but does not exist in the salivary duct, suggesting that oral bacteria might be involved in nitrate reduction (Kapil et al., 2020). This idea was supported by an experiment in which the use of an antibacterial mouthwash dramatically reduced nitrite levels in saliva (Tannenbaum et al., 1976). Various nitrate-reducing bacteria have been isolated from saliva, including *Veillonella*, *Lactobacillus*, *Micrococcus*, *Corynebacterium*, *Propionibacterium*, *Neisseria*, *Actinomyces*, commensal *Staphylococcus*, and *Rothia spp* (Doel et al., 2005). These bacteria are located in the deep crypts of the posterior part of the tongue, where they use nitrate in their respiration process and convert it into more reactive nitrites (Ma et al., 2018). There are two main pathways in bacteria for reducing nitrate. One is dissimilatory nitrate reduction to ammonium (DNRA), and the other is denitrification. The two pathways share a common initial step of reducing nitrate to nitrite by nitrate reductase. DNRA is a two-step reduction process that converts nitrate to ammonium (NH_4^+) by forming the intermediate nitrite (NO_2^-), which is completed by the

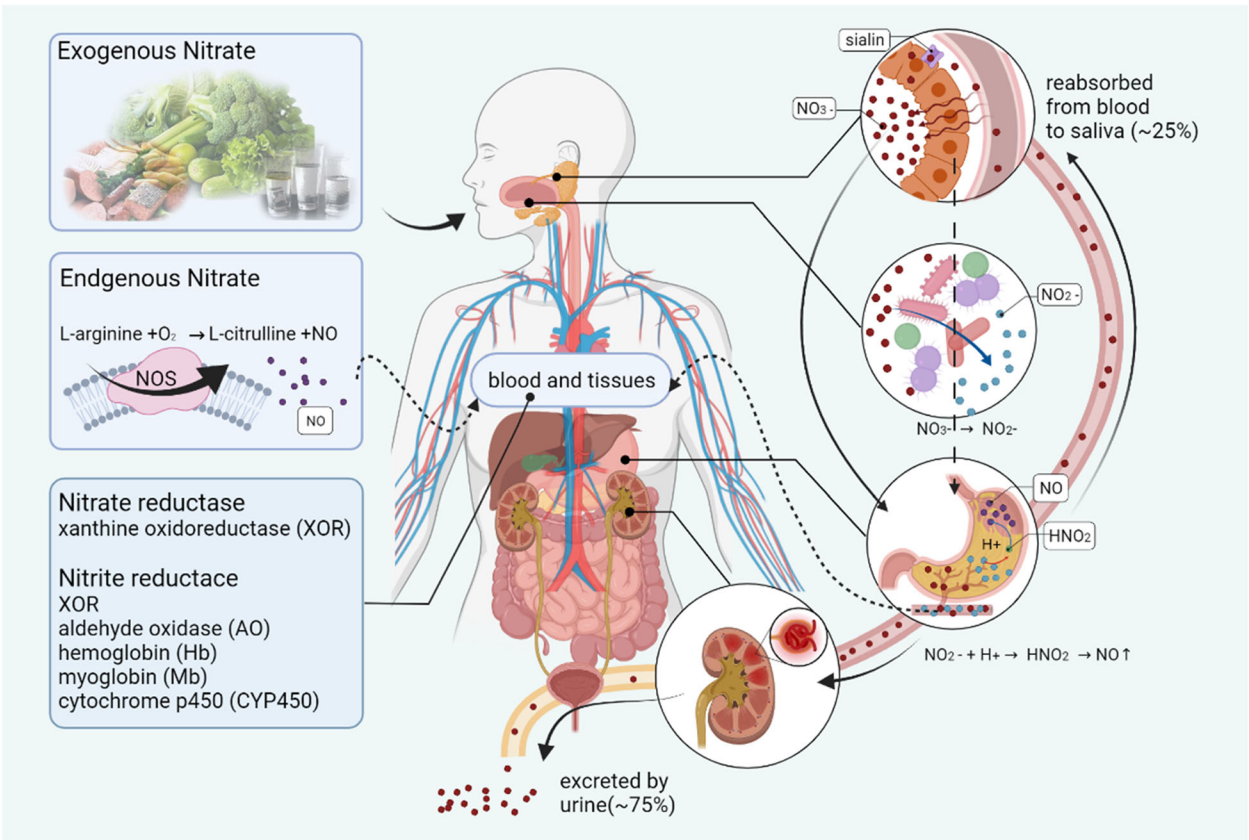

Fig. 1 Sources and metabolism of inorganic nitrate. There are two sources of nitrate in the human body: the endogenous production and the exogenous intake. The endogenous source of nitrate is the oxidation of NO synthesized by NOS. The exogenous source of nitrate is mainly the diet and green leafy vegetables take up more than 80% of the exogenous nitrate, in addition, drinking water and canned meat also contain nitrate, accounting for less than 20% of daily dietary intake. After ingestion, approximately 25% of the nitrate in the plasma is actively absorbed by the salivary glands. This process is also called entrosalivary circulation. Sialin, as a membrane transporter of nitrate, plays a significant role in this process. In addition, approximately two-thirds of the nitrate is excreted in the urine, and a small amount is excreted in the sweat and feces. Salivary nitrate is reduced to nitrite by nitrate-reducing-bacteria in oral cavity. The nitrite is then protonated into nitrous acid in stomach and immediately decomposed to derive NO. The fraction part of nitrite is transported by the circulatory system and reduced to NO in the blood and various systems, which process is well known as the nitrate-nitrite-NO pathway. Xanthine oxidoreductase (XOR) is the only nitrate reductase found in humans and mammals. In contrast, there are a series of nitrite reductase in human body, including XOR, aldehyde oxidase (AO), hemoglobin (Hb), myoglobin (Mb), cytochrome p450 (CYP450), complexes of the mitochondrial electron transport chain, vitamin C and even NOS

Nap/Nrf protein system where Nap is a cytosolic nitrate reductase, and Nrf is a nitrite reductase (Pandey et al., 2020; Sparacino-Watkins et al., 2014).

Salivary nitrite is then swallowed and protonated in the acidic gastric environment, generating nitrous acid (HNO_2) and further decomposing to NO and other nitrogen oxides locally (Lundberg et al., 1994). The remaining fraction of nitrite is systemically absorbed and then reduced to NO in the blood and tissues. There are various nitrite-reducing enzymes in different tissues, including XOR, aldehyde oxidase (AO), hemoglobin, myoglobin, cytochrome p450, complexes of the mitochondrial electron transport chain, vitamin C, and NOS (Kapil et al., 2020). Both XOR and AO are molybdenum-containing oxidases. XOR is widely distributed throughout the body, with the highest level of expression in the liver (Linder et al., 1999). Using NADH as the reducing substrate, XOR can reduce NO_2^- to NO at the molybdenum-binding site (Millar et al., 1998). XOR activity is increased at low oxygen tensions and pH values, demonstrating that XOR-mediated nitrite reduction may be a special case in ischemic situations, rather than a regular response under normal conditions (pH 7.4, normoxia) (Li et al., 2004). This suggests that the nitrate-nitrite-NO pathway is complementary to the classical NO pathway in special conditions, such as hypoxia and ischemia (Kapil et al., 2020) (Fig. 1).

3 Health-promotive functions of inorganic nitrate

3.1 Enhancing exercise performance

Exercise is often accompanied by a decrease in oxygen and pH levels in the muscles, which promotes the

nitrate-nitrite-NO pathway (Castello et al., 2006). Therefore, the role of nitrate in exercise has received considerable attention. Nitrate supplementation significantly reduced maximal oxygen consumption (VO_{2max}) in healthy volunteers under sub-maximum work rate (Larsen et al., 2007). Moreover, no difference in the amount of lactic acid in the blood was observed in this study, suggesting that nitrate did not increase energy production but enhanced muscle efficiency by improving the utilization of oxygen. These results are surprising because the O_2-cost of submaximal exercise is generally fixed and is not affected by other interventions such as physical age or health conditions (Suhr et al., 2013). This groundbreaking finding sparked widespread interest in nitrate, and numerous studies have since been conducted to verify whether nitrate can improve athletic performance. In a double-blind crossover experiment, nitrate-rich beetroot juice and blackcurrant juice with nearly zero nitrate content were administered to eight healthy young men. Then, a series of moderate- and high-intensity exercise tests were conducted over the next few days. Interestingly, nitrate reduced O_2 uptake during moderate-intensity exercise by 19% and delayed the attainment of the O_2-cost peak during high-intensity exercise (Bailey et al., 1985). Inorganic nitrate also delays fatigue and improves endurance in athletes during exercise. These beneficial effects have been observed in a variety of sports and exercises, including running, knee extension, desert marching, kayaking, and rowing (Jones et al., 2018). In addition, there is preliminary evidence that nitrate supplementation is conducive to post-exercise recovery (Clifford et al., 2016). These effects of nitrate are weakened in well-trained athletes, partly because of their higher resting plasma nitrate and nitrite levels (Arnold et al., 2015). The mechanism by which nitrate improves exercise performance remains unclear. It has been suggested that nitrate can reduce the energy cost of producing ATP and thus reduce the O_2-cost (Evangelista et al., 2010). Another hypothesis is that nitrate supplementation may ameliorate mitochondrial oxygenation and reduce proton leakage (Wilson, 1994). In addition, nitrate is associated with the upregulation of muscle calcium-processing capacity (Hernández et al., 2012) and boosting of blood flow, thus enhancing muscle contraction and oxygen delivery capacity (Ferguson et al., 2013).

3.2 Digestive system protection

Inorganic nitrate also plays a protective role in the digestive system. First, nitrate is reduced to nitrite by oral bacteria and then to NO by the gastric juice, thus enhancing the antimicrobial effect of gastric juice. Nitrite-rich saliva significantly increased NO levels in the stomach, thus suppressing *E. coli* and *C. albicans* numbers (Björne et al., 2006). On the other hand, by increasing gastric mucosal blood flow and mucus thickness, nitrate alleviates intestinal damage and ulceration caused by non-steroidal anti-inflammatory drugs and stress (Jädert et al., 2014; Jin et al., 2013). These effects were also likely mediated by NO, because no similar beneficial effect was observed in germ-free mice, indicating that nitrate-reducing bacteria were needed in this process. In addition, in the water-immersion restraint stress model, nitrate pretreatment (5 mmol/L $NaNO_3$) alleviated stress-induced gastric injury in rats by restoring gastric mucosal blood flow and NO levels. This result suggests that nitrate is a promising candidate for alleviating peptic ulcers under stressful conditions (Jin et al., 2013).

Salivary glands, as part of the digestive system, are also involved in the protective effects of nitrate. The secretion of nitrate by the salivary glands is associated with external stimuli; music was found to increase the saliva secretion of volunteers and promote the formation of nitrite in saliva (Jin et al., 2018). This mechanism may be beneficial under stressful conditions. The total amount of nitrate and nitrite in the saliva of volunteers increased significantly after bungee jumping, which was helpful in mitigating gastrointestinal damage after stress (Jin et al., 2013). In addition, nitrate can mediate the function of the salivary glands. In a rat model of hyposalivation induced by ovariectomy, 3 months of $NaNO_3$ treatment increased the salivary flow rate and improved the atrophy and fibrosis of the submandibular gland (Xu et al., 2018).

3.3 Alleviating senescence

Aging is often accompanied by a decrease in NO bioactivity, which is associated with a series of age-related diseases, including cardiovascular disease and metabolic dysfunction. Human studies have shown that nitrate levels decline with age (Toprakçi et al., 2000), suggesting endothelial function injury and a decrease in endogenous levels. Moreover, compared with younger volunteers, lower plasma nitrite concentrations were found in older subjects after dietary nitrate consumption, confirming a decline in exogenous NO bioavailability in the elderly. A decline in nitrate levels along with liver degeneration was observed in D-galactose-induced aged mice and naturally aged mice. Daily nitrate supplementation significantly restored nitrate levels in plasma and prevented cellular and structural senescence along with glucose- and lipid-metabolism degeneration in the liver. These findings suggest that inorganic nitrate has the potential to prevent senescence-related liver degeneration (Wang et al., 2018). Moreover, nitrate deficiency may accelerate aging and cause cardiovascular death. After up to 18 months of a low nitrite/nitrate diet, metabolic syndrome,

endothelial dysfunction, and cardiovascular death were observed in mice (Kina-Tanada et al., 2018).

3.4 Regulating microbial homeostasis

The gut and oral microflora are the most important microbiomes of the human body. The microbial environment is involved in the regulation of various physiological processes of the body, the imbalance of which may cause systemic dysfunction, including cardiovascular disease, diabetes, systemic infections, and even cancer (Sampaio-Maia et al., 2016; Sultan et al., 2021). Inorganic nitrate can regulate the homeostasis of microorganisms, thus producing a range of health-protective effects. In high-fat diet (HFD)-induced obesity, inorganic nitrate rebalances the gut microbiota and thus reduces obesity in mice. Nitrate enhances the abundance of *Bacteroidales* and *Alistipes (*Ma et al., 2020*)*, which are reported to have protective effects against HFD-induced non-alcoholic fatty liver disease (Tang et al., 2018). Disorder of gut microbiota homeostasis may also lead to inflammatory bowel disease. In mice loaded with dextran sodium sulfate, nitrate pretreatment increased the abundance of *Lactobacillus*, *Ruminococcaceae*, and *Prevotellaceae*, improved colon length, prevented apoptosis of colonic epithelial cells, and alleviated inflammatory cell infiltration in the colon and peripheral blood (Hu et al., 2020). Inorganic nitrate appears to also play a protective role in nonsteroidal anti-inflammatory drug-induced injury by maintaining microbiota homeostasis. Another study showed that dietary nitrate rebalances gut microbiome dysbiosis and partially prevents colon injury after total body irradiation (Wang et al., 2020).

Inorganic nitrate is also involved in the regulation of oral flora, which can have beneficial effects on systemic health. Nitrate supplementation in drinking water significantly altered the flora in the saliva of healthy volunteers, and the abundance of nitrate-reducing bacteria was markedly increased (Sinha et al., 2021). These changes may have additional effects; a larger nitrate-reducing bacteria population can accelerate the nitrate-nitrite-NO pathway and promote the NO-like benefit effect of nitrate. Nitrate treatment upregulated the proportion of *Rothia* and *Neisseria* and reduced blood pressure in healthy elderly participants (Vanhatalo et al., 2018) (Fig. 2).

4 Disease prevention functions of inorganic nitrate

4.1 Improving cardiovascular function

Organic nitrate drugs are widely used in the therapy of hypertension, and their effects mainly depend on the production of NO. As a supplementary source of NO, the antihypertensive effect of inorganic nitrate has received increasing attention. This effect of nitrate has been verified in a series of animal models of hypertension. In a hypertension model of rats with mononephrectomy and a high-salt diet, supplementation with $NaNO_3$ not only lowered blood pressure, but also alleviated cardiac and renal fibrosis (Carlström et al., 2011). Similarly, in another model of salt-induced hypertension in Dahl salt-sensitive rats, nitrate reduced the effect of salt loading on mean arterial pressure by 60% (Morris Jr. et al., 2019). The hypotensive effect of nitrate was also observed in rats with high fructose-induced metabolic syndrome (Essawy et al., 2014). In elderly adults with prehypertension, systolic blood pressure (SBP) was reduced by 8 mmHg after 4 weeks of nitrate intake (Rammos et al., 2014). After a single dose of dietary nitrate via beetroot juice (3.3 mmol/205 mg NO_3^-), the average blood pressure of 15 patients with hypertension decreased by 11/10 mmHg (Ghosh et al., 2013). Surprisingly, the hypotensive effects of nitrates do not conflict with those of traditional antihypertensive drugs. Beetroot juice with nitrate (6.4 mmol/397 mg) significantly lowered clinical, home, and 24 h ambulatory blood pressure in 32 hypertensive patients with and without medication (Kapil et al., 2015).

The anti-hypertensive effect of dietary nitrate has also been observed in healthy people. A meta-analysis of 34 studies including healthy subjects and patients with hypertension indicated that inorganic nitrate intake significantly reduced resting SBP by 4.80 mmHg and resting DBP by 1.74 mmHg. Compared with healthy people, inorganic nitrate showed a more significant antihypertensive effect in patients with hypertension. (Jackson et al., 2018). There is a explanation that exogenous nitrate seemed to be more effective when NO availability was reduced. A single dose of dietary nitrate (3.3 mmol (205 mg) via beetroot juice reduced peak blood pressure by 11/10 mmHg in untreated hypertensive patients (Ghosh et al., 2013). The same hypotensive effect could be replicated in healthy volunteers only after a higher concentration (22.5 mmol) of nitrate was administered via beetroot juice (Webb et al., 2008). These results suggest that hypertensive patients have a higher sensitivity to inorganic nitrates, which means a lower threshold for anti-hypertensive effects and a subsequent greater drop in blood pressure (Kapil et al., 2020). The mainstream viewpoint is that the antihypertensive effect of nitrate is derived from the vasodilatation of NO. Cyclic guanosine monophosphate (cGMP) is a sensitive marker of NO production and bioactivity, and its concentration in the circulation significantly increases after nitrate supplementation, which indicates that the antihypertensive effect is induced by NO (Jackson et al., 2018). Another study suggested that increased sensitivity to nitrate in a model of spontaneous hypertension may be associated with increased XOR expression (Laakso et al., 1998).

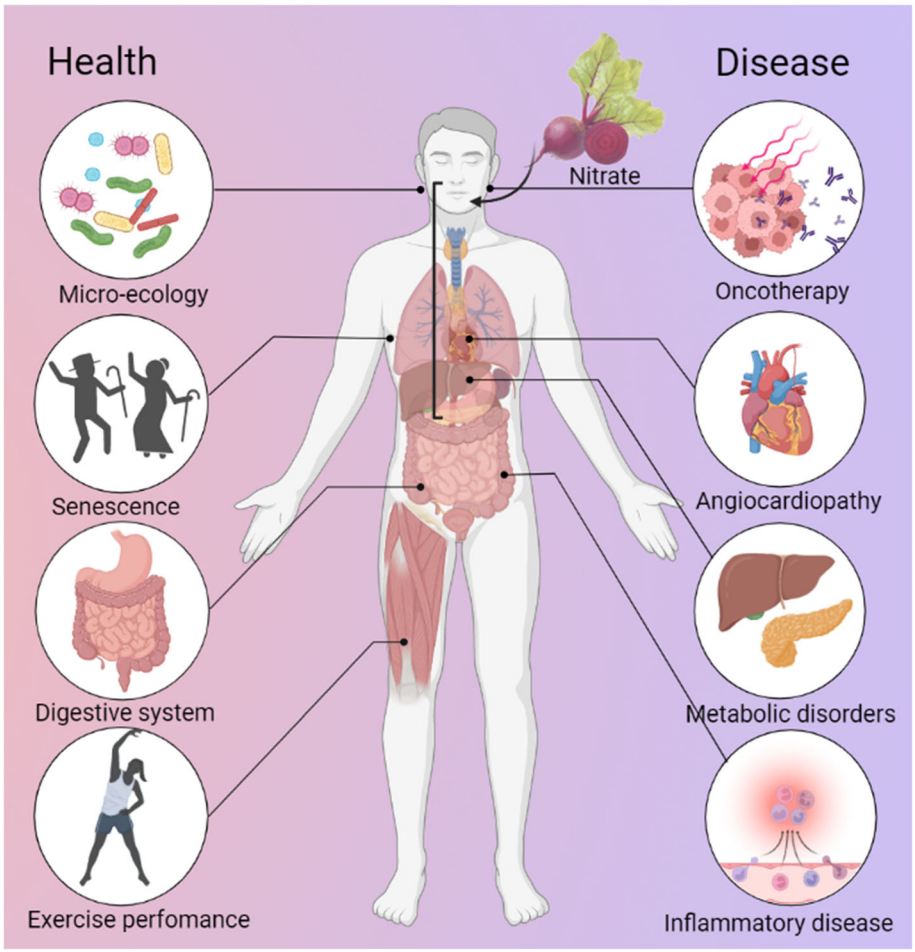

Fig. 2 Health promotion and disease prevention functions of inorganic nitrate. Inorganic nitrate has shown beneficial physiological effects in multiple organs and systems. On the one hand, nitrate has a series of beneficial functions in health promotion. Nitrate can regulate microbial homeostasis, delay senescence, protect digestive system, and enhance exercise performance. On the other hand, nitrate performs physiological functions in various systemic diseases, including assist tumor treatments, relieve cardiovascular diseases, alleviate metabolic disorders, and mitigate inflammatory conditions

NO deficiency is an important cause of both heart failure with reduced ejection fraction (HFrEF) and heart failure with preserved ejection fraction (HFpEF) (van Heerebeek et al., 2012). Nitrate and nitrite can play NO-like roles and rescue cardiovascular function under conditions of NO deficiency (Paulus & Tschöpe, 2013). Exogenous nitrite application has shown beneficial effects in the hearts of many animals, including fish, amphibians, and mammals. In these animals, nitrite improved systolic and diastolic function, reduced left ventricular pressure, and increased both stroke volume and work per stroke (Angelone et al., 2012; Pellegrino et al., 2009). Dietary nitrates showed similar benefits. In a mouse model of doxorubicin-induced cardiac dysfunction, $NaNO_3$ administration improved left ventricular systolic pressure, end-diastolic pressure, and ejection fraction of mice (Zhu et al., 2011). In addition, beetroot juice rich in nitrate (1 mmol/kg) increased the resting blood flow and vascular conductivity of rats with heart failure by 22% and 20%, respectively, and the improvement was more obvious during exercise (Ferguson et al., 1985). Similarly, dietary nitrate has shown beneficial effects during exercise in patients with heart failure. Maximum muscle speed and strength during exercise increased in heart failure patients after ingestion of nitrate-concentrated beetroot juice (Coggan et al., 2015). In addition, there is also evidence that inorganic nitrate intake maintains ejection fraction during exercise (Zamani et al., 2015) and enhances exercise tolerance in elderly patients with heart failure (Eggebeen et al., 2016). This result suggests that inorganic nitrate may be a promising candidate for improving the quality of life of patients with heart failure.

NO can reduce the adhesion, aggregation, recruitment, and formation of platelet-leukocyte aggregates (Chung et al., 2004; Radomski et al., 1987; Radomski et al.,

1990). Inhibition of platelet activity by inorganic nitrites and nitrates was also observed. $NaNO_2$ and $NaNO_3$ pretreatment inhibited platelet aggregation and prolonged bleeding time. In the same study, opposite results were observed in mice fed a low-nitrate diet (Park et al., 2013). In patients with endothelial dysfunction and mild hypercholesterolemia, 6 weeks of dietary nitrate supplementation reduced platelet p-selectin expression and leukocyte-platelet aggregation, which contributed to reducing thrombosis (Velmurugan et al., 2016).

4.2 Regulating metabolic diseases

Inorganic nitrate may be involved in the regulation of metabolic diseases. A long-term nitrate-reduced diet resulted in a series of metabolic abnormalities in C57BL6 mice, including increased visceral obesity, dyslipidemia, decreased glucose tolerance, and insulin resistance (Kina-Tanada et al., 2018). In mice with eNOS deficiency, 8–10 weeks of $NaNO_3$ supplementation was associated with less body fat and weight loss in the mice, as well as improved glucose homeostasis (Carlström et al., 2010). In another study, acute nitrate ingestion improved insulin resistance and glucose clearance in adenosine A2B-deficient mice (Peleli et al., 2015). In addition, several independent studies have shown that dietary nitrates can improve glucose tolerance and insulin sensitivity (Essawy et al., 2014; Gheibi et al., 2017; Khalifi et al., 2015; Li et al., 2016). There is also evidence that inorganic nitrate promotes the conversion of white adipose tissue to brown adipose tissue by activating the nitrate-nitrite-NO pathway (Roberts et al., 2015). Data suggests that nitrate plays an important role in metabolism and may prevent diabetes and obesity. However, these beneficial effects have not been replicated in human trials. Dietary nitrate intake for up to 2 weeks did not improve insulin sensitivity in patients with type 2 diabetes (Gilchrist et al., 2014). The absence of a beneficial function of nitrate in patients with type 2 diabetes may be due to the use of metformin (Cabreiro et al., 2013; Forslund et al., 2015). A recent review also suggested that the difference in ascorbic acid metabolism between humans and rodents may explain the lack of beneficial effects of inorganic nitrate metabolism in patients with type 2 diabetes (Bahadoran et al., 2021).

4.3 Alleviating inflammatory conditions

Inflammation and oxidative stress are the main causes of ischemia-reperfusion (IR) injury, and the protective effect of nitrite against this has been observed in a series of animal models. Nitrite injection prior to ligation effectively reduces the size of the cardiac infarction in vivo (Duranski et al., 2005; Gonzalez et al., 2008). Similar protective effects have been observed in the study of nitrate in different organs. In a study of renal IR injury in mice, $NaNO_3$ administration for 2 weeks prior to ischemic injury reserved the glomerular filtration rate and renal plasma flow (Yang et al., 2017). In addition, dietary nitrate pretreatment alleviated hypoxia-induced mitochondrial dysfunction and oxidative stress in rats by upregulating L-arginine levels in tissues and reducing cardiac arginase expression (Ashmore et al., 2014). In a recent study, nitrate pretreatment alleviated hepatic IR injury in mice by activating the NRF2 pathway and modulating oxidative stress (Li et al., 2021a). In healthy volunteers, temporarily blocking forearm blood flow caused transient endothelial dysfunction in the forearm and significantly reduced brachial artery flow-mediated dilation (by 60%). The damage was prevented in volunteers who had been given nitrates beforehand, suggesting that nitrate may prevent IR injury in humans (Webb et al., 2008).

Dietary nitrates can induce anti-inflammatory effects in a variety of conditions by modulating the levels of inflammatory factors. In a mouse model of atherosclerosis, inorganic nitrate supplementation increased the levels of IL-10, an anti-inflammatory factor, in plaques (Bakker et al., 2016), and there was also evidence that inorganic nitrate supplementation was associated with an increased proportion of collagen expression in plaques, suggesting that nitrate may be effective in reducing inflammation within plaques (Stefanadis et al., 2017). In another study of diabetic rats, after $NaNO_3$ supplementation for 8 weeks, the expression of iNOS was decreased in the soleus muscle and epididymal adipose tissue (Gheibi et al., 2018). Moreover, dietary ingestion of cooked or raw beetroot for 2 weeks was associated with a decline in systemic inflammation and biomarkers of endothelial dysfunction in healthy individuals, including ICAM-1, VCAM-1, E-selectin, IL-6, hsCRP, and TNF-α (Asgary et al., 2016). The protective effect of nitrate through the inhibition of leukocyte recruitment has been demonstrated in a variety of inflammatory models. For example, CXCL2 perfusion can induce leukocyte recruitment and promote leukocyte adhesion and migration. These effects were alleviated by intravenous injection of $NaNO_2$ 1 h before CXCL2 perfusion. In the same study, a one-week treatment with $NaNO_3$ (10 mmol/L) in drinking water also reduced baseline leukocyte rolling and reduced leukocyte adhesion and migration induced by CXCL2 perfusion (Jädert et al., 2012). In addition, the anti-leukocyte effects of nitrate were also observed in sodium glucan sulfate-induced colitis in rats (Jädert et al., 2014; Ohtake et al., 2010), intestinal obstruction in mice (Cosyns et al., 2015), and chlorine-induced lung injury in rabbits (Samal et al., 2012).

4.4 Preventing post-radiation injury

Radiotherapy and chemotherapy are commonly used as adjuvant therapies for cancer, but their side effects may

seriously affect quality of life; reducing the side effects of treatment has always been a major challenge in the treatment of cancer. Reactive oxygen species (ROS) and oxidative stress are important factors in radiation injury (Tominaga et al., 2004). Evidence suggests that inorganic nitrates help mitigate radiation damage by reducing oxidative stress. In a mouse model of total body irradiation, nitrate in drinking water effectively restored the levels of red blood cells, leukocytes, and platelets in the peripheral blood by lowering ROS levels and reducing apoptosis (Chang et al., 2019). In addition, nitrate supplementation reduced the decline in salivary flow rate by preventing damage to acinar cells and microvascular endothelial cells, and alleviated weight loss induced by IR. Nitrate has been suggested to inhibit pyroapoptosis mediated by the NLPR3 inflammasome, and to reduce ROS production, thus maintaining mitochondrial homeostasis (Li et al., 2021b). The protective effect of nitrate has also been confirmed in larger mammals and in human parotid gland cells. Nitrate increased the expression of sialin in the parotid glands of miniature pigs, which could facilitate nitrate influx into cells, promote the regeneration of salivary gland cells, and reduce apoptosis (Feng et al., 2021a). In addition to its protective effects in radiation therapy, nitrate also has benefits in chemotherapy. Cisplatin is used as a chemotherapeutic drug for oral squamous cell carcinoma (OSCC). However, the activation of REDD1 and AKT can suppress the sensitivity of tumor cells, thus reducing the efficacy of cisplatin. In a recent study, inorganic nitrate reduced the expression of REDD1 and suppressed the activation of AKT signaling, thus increasing the sensitivity of OSCC cells to cisplatin (Feng et al., 2021b). These beneficial effects suggest that inorganic nitrates hold good potential in alleviating the side effects of radiotherapy and improving the quality of life of patients (Fig. 2).

5 Mechanisms of beneficial effects of inorganic nitrate

NO is an important gaseous signaling molecule that is involved in the regulation of many physiological processes, including the stimulation of reparative angiogenesis and reduction of oxidative stress (Gluvic et al., 2020; Sengupta et al., 2004; Sessa, 2009). The classical pathway of NO synthesis is mediated by NOS, which is aerobic. Reduction in NO bioavailability or bioactivity is associated with a variety of systemic diseases. The nitrate-nitrite-NO pathway is a powerful parallel pathway to the classical NO pathway and can be seen as a backup system to ensure continued NO supply when the O_2-dependent NOS may be dysfunctional. In other words, under pathological conditions dominated by regional and systemic ischemia, nitrate and nitrite in food can serve as an effective reserve source of NO (Bryan & Ivy, 2015).

The free radical nature of NO is the basis for its role in physiological signaling (Weitzberg & Lundberg, 2013; Lundberg et al., 2015). Guanylyl cyclase (GC) is one of the main intracellular receptors of NO. NO can activate GC in various cell types, such as smooth muscle cells, circulating platelets, and leukocytes (Ghimire et al., 2017). Activated soluble GC further enhances the production of the second messenger cGMP, which in turn relaxes smooth muscle cells and inhibits adhesion of platelets and leukocytes (Ghimire et al., 2017; Carlström et al., 2018). NO also controls multiple intracellular functions, such as mitochondrial function, ROS production, and other signal transduction pathways (Ghimire et al., 2017). Research has shown that through the nitrate-nitrite-NO pathway, nitrate and nitrite can also activate GC-cGMP signaling and play an NO-like role in several physiological and pathological conditions (Raubenheimer et al., 2019). GC-1 inhibitors block the vasodilation of nitrite in rodents and pigs, suggesting that the vasodilation of nitrite may be mediated by the NO-GC pathway (Botden et al., 2012).

With the development of numerous studies, there is increasing evidence showing that nitrate has a protective effect on salivary glands and other organs by regulating the expression of sialin. Under hypoxic or acidic conditions, NO synthesis primarily occurs via the nitrate-nitrite-NO exogenous pathway. Remarkably, irradiation induced a hypoxic and acidic environment in salivary glands. By inducing a prolonged increase in blood flow (Lundberg et al., 2008) and by increasing gland microvascularization, dietary nitrate supplementation resulted in NO production and reduced hypoxia, which was beneficial for the production of saliva by acinar cells. In addition, nitrate-mediated NO formation also increased sialin expression and upregulated the EGFR-AKT-MAPK signaling pathway (Feng et al., 2021a). These classical pathways are responsible for promoting cell proliferation, maintaining cell survival, and preventing apoptosis (Sabbah et al., 2020; Sun et al., 2015; Zhang et al., 2011). A recent study demonstrated a nitrate-sialin feedback loop in vivo, using a miniature pig model, and in vitro, using human parotid gland cells (hPGCs); nitrate increased sialin expression, and sialin facilitated nitrate influx into cells. The study showed that sialin-mediated nitrate transport plays a critical role in maintaining the proliferation and survival of parotid gland epithelial cells and preventing IR damage and that sialin acts as a nitrate transporter. Preventive nitrate administration increases sialin expression, promotes acinar and ductal cell proliferation, and reduces apoptosis via the EGFR-AKT-MAPK signaling pathway (Feng et al., 2021a) (Fig. 3).

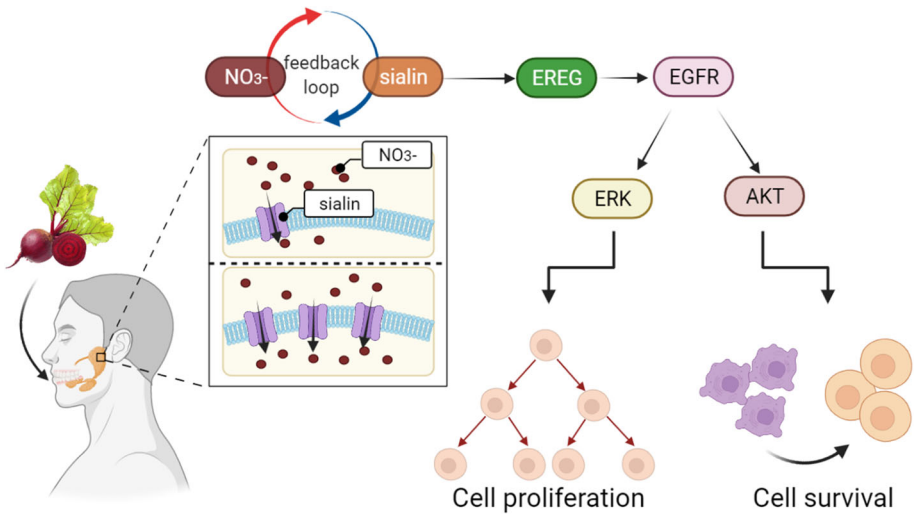

Fig. 3 The protective effect of nitrate by regulation of sialin. Inorganic nitrate has a protective effect on salivary glands and other organs by regulating the expression of sialin. Dietary nitrate supplementation increases sialin expression and upregulates EGFR-AKT-MAPK signaling pathway. These classical pathways are responsible for promoting cell proliferation, maintaining cell survival, and preventing apoptosis. In addition, the upregulation of sialin boosts nitrate influx into cells, indicating that there is a positive feedback loop between nitrate and sialin, which can further promote the protective effect of nitrate

S-nitrosothiols can be obtained by nitrosation of thiol groups, such as cysteine residues in proteins. S-nitrosation is considered a mechanism of post-translational dynamic regulation of proteins (Evangelista et al., 2010). Evidence shows that nitrite can effectively induce heme nitrosylation and S-nitrosation in mammals to participate in signal regulation in a different way from NO (Kapil et al., 2020). In addition, nitrate derived S-nitrosothiol formation can modulate inflammation, reduce reactive oxygen species formation by inhibiting mitochondrial respiration, and activate cyclic GMP-dependent signaling under anoxia (Lundberg et al., 2008).

6 Advantages and safety of inorganic nitrate

There are numerous advantages of inorganic nitrate compared with traditional organic nitrate drugs. On the one hand, there are some concerns about the use of organic nitrates, such as side effects of headache and postural hypotension (Tarkin & Kaski, 2016). In contrast, inorganic nitrates are less likely to cause symptomatic hypotension or syncope (Kapil et al., 2020). In addition, long-term use of organic nitrates not only leads to a decline in potency, but may also lead to endothelial dysfunction and increased long-term cardiovascular risk (Tarkin & Kaski, 2016). These side effects were not observed upon treatment with inorganic nitrates/nitrites; conversely, the potential to restore NO availability and improve endothelial dysfunction has been reported (Paulus & Tschöpe, 2013). Moreover, the antihypertensive effect of nitrate was observed even after 1 year of inorganic nitrate supplementation, proving that inorganic nitrate has no tolerance effect like organic nitrate (Münzel et al., 2005).

In terms of the safety of inorganic nitrate, there is concern about nitrate-derived nitrite in the metabolic process. Hemoglobin can be oxidized to methemoglobin by nitrite, thus causing methemoglobinemia. Nevertheless, current evidence suggests that nitrate supplementation is not associated with elevated metHb levels (Kapil et al., 2015; Velmurugan et al., 2016). Another concern about the safety of nitrates is that in the acidic environment of the stomach, nitrates may produce nitrosamines, which have been reported to be associated with esophageal, gastric, colon, and other cancers (Bedale et al., 2016). Nitrite from dietary intake or reduction by oral bacteria can be protonated into nitrous acid in the stomach and produce highly oxidizing nitrous oxide, which can further nitrosate secondary or tertiary amines and eventually form nitrosamines (Kapil et al., 2020). However, a report by the International Agency for Research on Cancer suggests that there is insufficient evidence to link dietary nitrate to overall cancer (IARC monographs on the evaluation of carcinogenic risks to humans, 2010). Actually, the nitrate/nitrite concentrations used for treatment are far below the risk level, and nitrosamine production is inappreciable enough to ensure negligible carcinogenic risk (Lundberg et al., 2008). Moreover, dietary nitrates seem to play a protective role in the risk of stomach cancer (IARC monographs on the evaluation of carcinogenic risks to humans, 2010). Nitrates are highly concentrated in green leafy vegetables

and vegetable-rich diets. Vegetable components such as polyphenols, vitamin C, and vitamin E can inhibit the formation of carcinogenic nitrosamines, suggesting that dietary nitrate supplementation through vegetables is a safe method (Milton-Laskibar et al., 2021).

7 Conclusion

In recent years, an increasing number of studies have shown that nitrate plays an important physiological role in the human body. As a part of the daily diet, inorganic nitrate is mainly consumed in green leafy vegetables. Nitrate not only affects enzymatic NO, but also plays important physiological roles through the non-enzymatic NO pathway. Inorganic nitrate has been shown to be an exogenous alternative to biological NO. Dietary nitrate has potential positive effects on health and disease, including improving muscle function, maintaining microbial homeostasis, regulating vascular tension, antioxidizing, inhibiting the release of inflammatory factors, and regulating glucose metabolism. Sialin, functioning as a nitrate transporter, plays an indispensable role in various physiological functions. Nitrate-sialin may provide the biological basis for homeostasis medicine in health and diseases. More biological functions and mechanisms of nitrate remain to be elucidated. Inorganic nitrate research is still in the pre-clinical stage, and nitrate has not been used as the main treatment method at present. The mechanism and effective dose of nitrate therapy are not entirely clear. Through reasonable design of dosage form, the beneficial effect of nitrate can be utilized to a greater extent. With further research, nitrate, as an easily available nutrient, is a promising candidate for health promotion and disease prevention.

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References

Angelone T, Gattuso A, Imbrogno S, Mazza R, Tota B. Nitrite is a positive modulator of the frank-Starling response in the vertebrate heart. Am J Phys Regul Integr Comp Phys. 2012;302(11):R1271–81. https://doi.org/10.1152/ajpregu.00616.2011.

Arnold JT, Oliver SJ, Lewis-Jones TM, Wylie LJ, Macdonald JH. Beetroot juice does not enhance altitude running performance in well-trained athletes. Appl Physiol Nutr Metab. 2015;40(6):590–5. https://doi.org/10.1139/apnm-2014-0470.

Asgary S, Afshani MR, Sahebkar A, Keshvari M, Taheri M, Jahanian E, et al. Improvement of hypertension, endothelial function and systemic inflammation following short-term supplementation with red beet (*Beta vulgaris* L.) juice: a randomized crossover pilot study. J Hum Hypertens. 2016. https://doi.org/10.1038/jhh.2016.34.

Ashmore T, Fernandez BO, Branco-Price C, West JA, Cowburn AS, Heather LC, et al. Dietary nitrate increases arginine availability and protects mitochondrial complex I and energetics in the hypoxic rat heart. J Physiol. 2014;592(21):4715–31. https://doi.org/10.1113/jphysiol.2014.275263.

Bahadoran Z, Mirmiran P, Kashfi K, Ghasemi A. Lost-in-translation of metabolic effects of inorganic nitrate in type 2 diabetes: is ascorbic acid the answer? Int J Mol Sci. 2021;22(9). https://doi.org/10.3390/ijms22094735.

Bailey SJ, Winyard P, Vanhatalo A, Blackwell JR, Dimenna FJ, Wilkerson DP, et al. Dietary nitrate supplementation reduces the O_2 cost of low-intensity exercise and enhances tolerance to high-intensity exercise in humans. J Appl Physiol. 1985;2009(4):1144–55. https://doi.org/10.1152/japplphysiol.00722.2009.

Bakker JR, Bondonno NP, Gaspari TA, Kemp-Harper BK, McCashney AJ, Hodgson JM, et al. Low dose dietary nitrate improves endothelial dysfunction and plaque stability in the ApoE(−/−) mouse fed a high fat diet. Free Radic Biol Med. 2016;99:189–98. https://doi.org/10.1016/j.freeradbiomed.2016.08.009.

Bedale W, Sindelar JJ, Milkowski AL. Dietary nitrate and nitrite: benefits, risks, and evolving perceptions. Meat Sci. 2016;120:85–92. https://doi.org/10.1016/j.meatsci.2016.03.009.

Björne H, Weitzberg E, Lundberg JO. Intragastric generation of antimicrobial nitrogen oxides from saliva--physiological and therapeutic considerations. Free Radic Biol Med. 2006;41(9):1404–12. https://doi.org/10.1016/j.freeradbiomed.2006.07.020.

Blekkenhorst LC, Prince RL, Ward NC, Croft KD, Lewis JR, Devine A, et al. Development of a reference database for assessing dietary nitrate in vegetables. Mol Nutr Food Res. 2017;61(8). https://doi.org/10.1002/mnfr.201600982.

Botden IP, Batenburg WW, de Vries R, Langendonk JG, Sijbrands EJ, Danser AH. Nitrite- and nitroxyl-induced relaxation in porcine coronary (micro-) arteries: underlying mechanisms and role as endothelium-derived hyperpolarizing factor(s). Pharmacol Res. 2012;66(5):409–18. https://doi.org/10.1016/j.phrs.2012.07.006.

Bryan NS, Ivy JL. Inorganic nitrite and nitrate: evidence to support consideration as dietary nutrients. Nutr Res. 2015;35(8):643–54. https://doi.org/10.1016/j.nutres.2015.06.001.

Cabreiro F, Au C, Leung KY, Vergara-Irigaray N, Cochemé HM, Noori T, et al. Metformin retards aging in *C. elegans* by altering microbial folate and methionine metabolism. Cell. 2013. https://doi.org/10.1016/j.cell.2013.02.035.

Carlström M, Larsen FJ, Nyström T, Hezel M, Borniquel S, Weitzberg E, et al. Dietary inorganic nitrate reverses features of metabolic syndrome in endothelial nitric oxide synthase-deficient mice. Proc Natl Acad Sci U S A. 2010;107(41):17716–20. https://doi.org/10.1073/pnas.1008872107.

Carlström M, Lundberg JO, Weitzberg E. Mechanisms underlying blood pressure reduction by dietary inorganic nitrate. Acta Physiol (Oxford). 2018;224(1). https://doi.org/10.1111/apha.13080.

Carlström M, Persson AE, Larsson E, Hezel M, Scheffer PG, Teerlink T, et al. Dietary nitrate attenuates oxidative stress, prevents cardiac and renal injuries, and reduces blood pressure in salt-induced hypertension. Cardiovasc Res. 2011; 89(3):574–85. https://doi.org/10.1093/cvr/cvq366.

Castello PR, David PS, McClure T, Crook Z, Poyton RO. Mitochondrial cytochrome oxidase produces nitric oxide under hypoxic conditions: implications for oxygen sensing and hypoxic signaling in eukaryotes. Cell Metab. 2006;3(4): 277–87. https://doi.org/10.1016/j.cmet.2006.02.011.

Chang S, Hu L, Xu Y, Li X, Ma L, Feng X, et al. Inorganic nitrate alleviates total body irradiation-induced systemic damage by decreasing reactive oxygen species levels. Int J Radiat Oncol Biol Phys. 2019;103(4):945–57. https://doi.org/10.1016/j.ijrobp.2018.11.021.

Chung AW, Radomski A, Alonso-Escolano D, Jurasz P, Stewart MW, Malinski T, et al. Platelet-leukocyte aggregation induced by PAR agonists: regulation by nitric oxide and matrix metalloproteinases. Br J Pharmacol. 2004;143(7):845–55. https://doi.org/10.1038/sj.bjp.0705997.

Clifford T, Bell O, West DJ, Howatson G, Stevenson EJ. The effects of beetroot juice supplementation on indices of muscle damage following eccentric exercise. Eur J Appl Physiol. 2016;116(2):353–62. https://doi.org/10.1007/s00421-015-3290-x.

Coggan AR, Leibowitz JL, Spearie CA, Kadkhodayan A, Thomas DP, Ramamurthy S, et al. Acute dietary nitrate intake improves muscle contractile function in patients with heart failure: a double-blind, placebo-controlled, randomized trial. Circ Heart Fail. 2015;8(5):914–20. https://doi.org/10.1161/circheartfailure.115.002141.

Cosyns SM, Shiva S, Lefebvre RA. Protective effect of exogenous nitrite in postoperative ileus. Br J Pharmacol. 2015;172(20):4864–74. https://doi.org/10.1111/bph.13255.

Crawford JH, Chacko BK, Pruitt HM, Piknova B, Hogg N, Patel RP. Transduction of NO-bioactivity by the red blood cell in sepsis: novel mechanisms of vasodilation during acute inflammatory disease. Blood. 2004;104(5):1375–82. https://doi.org/10.1182/blood-2004-03-0880.

Doel JJ, Benjamin N, Hector MP, Rogers M, Allaker RP. Evaluation of bacterial nitrate reduction in the human oral cavity. Eur J Oral Sci. 2005;113(1):14–9. https://doi.org/10.1111/j.1600-0722.2004.00184.x.

Duncan C, Dougall H, Johnston P, Green S, Brogan R, Leifert C, et al. Chemical generation of nitric oxide in the mouth from the enterosalivary circulation of dietary nitrate. Nat Med. 1995;1(6):546–51. https://doi.org/10.1038/nm0695-546.

Duranski MR, Greer JJ, Dejam A, Jaganmohan S, Hogg N, Langston W, et al. Cytoprotective effects of nitrite during in vivo ischemia-reperfusion of the heart and liver. J Clin Invest. 2005;115(5):1232–40. https://doi.org/10.1172/jci22493.

Eggebeen J, Kim-Shapiro DB, Haykowsky M, Morgan TM, Basu S, Brubaker P, et al. One week of daily dosing with beetroot juice improves submaximal endurance and blood pressure in older patients with heart failure and preserved ejection fraction. JACC Heart Fail. 2016;4(6):428–37. https://doi.org/10.1016/j.jchf.2015.12.013.

Essawy SS, Abdel-Sater KA, Elbaz AA. Comparing the effects of inorganic nitrate and allopurinol in renovascular complications of metabolic syndrome in rats: role of nitric oxide and uric acid. Arch Med Sci. 2014;3(3):537–45. https://doi.org/10.5114/aoms.2013.33222.

Evangelista AM, Rao VS, Filo AR, Marozkina NV, Doctor A, Jones DR, et al. Direct regulation of striated muscle myosins by nitric oxide and endogenous nitrosothiols. PLoS One. 2010;5(6):e11209. https://doi.org/10.1371/journal.pone.0011209.

Feng X, Wu Z, Xu J, Xu Y, Zhao B, Pang B, et al. Dietary nitrate supplementation prevents radiotherapy-induced xerostomia. Elife. 2021a;10. https://doi.org/10.7554/eLife.70710.

Feng Y, Cao X, Zhao B, Song C, Pang B, Hu L, et al. Nitrate increases cisplatin chemosensitivity of oral squamous cell carcinoma via REDD1/AKT signaling pathway. Sci China Life Sci. 2021b;64(11):1814–28. https://doi.org/10.1007/s11427-020-1978-4.

Ferguson SK, Hirai DM, Copp SW, Holdsworth CT, Allen JD, Jones AM, et al. Effects of nitrate supplementation via beetroot juice on contracting rat skeletal muscle microvascular oxygen pressure dynamics. Respir Physiol Neurobiol. 2013;187(3):250–5. https://doi.org/10.1016/j.resp.2013.04.001.

Ferguson SK, Holdsworth CT, Colburn TD, Wright JL, Craig JC, Fees A, et al. Dietary nitrate supplementation: impact on skeletal muscle vascular control in exercising rats with chronic heart failure. J Appl Physiol. 1985;2016(3):661–9. https://doi.org/10.1152/japplphysiol.00014.2016.

Forslund K, Hildebrand F, Nielsen T, Falony G, Le Chatelier E, Sunagawa S, et al. Disentangling type 2 diabetes and metformin treatment signatures in the human gut microbiota. Nature. 2015;528(7581):262–6. https://doi.org/10.1038/nature15766.

Gassara F, Kouassi AP, Brar SK, Belkacemi K. Green alternatives to nitrates and nitrites in meat-based products-a review. Crit Rev Food Sci Nutr. 2016;56(13): 2133–48. https://doi.org/10.1080/10408398.2013.812610.

Gheibi S, Bakhtiarzadeh F, Jeddi S, Farrokhfall K, Zardooz H, Ghasemi A. Nitrite increases glucose-stimulated insulin secretion and islet insulin content in obese type 2 diabetic male rats. Nitric Oxide. 2017;64:39–51. https://doi.org/10.1016/j.niox.2017.01.003.

Gheibi S, Jeddi S, Carlström M, Gholami H, Ghasemi A. Effects of long-term nitrate supplementation on carbohydrate metabolism, lipid profiles, oxidative stress, and inflammation in male obese type 2 diabetic rats. Nitric Oxide. 2018;75:27–41. https://doi.org/10.1016/j.niox.2018.02.002.

Ghimire K, Altmann HM, Straub AC, Isenberg JS. Nitric oxide: what's new to NO? Am J Phys Cell Phys. 2017;312(3):C254–62. https://doi.org/10.1152/ajpcell.00315.2016.

Ghosh SM, Kapil V, Fuentes-Calvo I, Bubb KJ, Pearl V, Milsom AB, et al. Enhanced vasodilator activity of nitrite in hypertension: critical role for erythrocytic xanthine oxidoreductase and translational potential. Hypertension. 2013;61(5): 1091–102. https://doi.org/10.1161/hypertensionaha.111.00933.

Gilchrist M, Winyard PG, Fulford J, Anning C, Shore AC, Benjamin N. Dietary nitrate supplementation improves reaction time in type 2 diabetes: development and application of a novel nitrate-depleted beetroot juice placebo. Nitric Oxide. 2014;40:67–74. https://doi.org/10.1016/j.niox.2014.05.003.

Gluvic ZM, Obradovic MM, Sudar-Milovanovic EM, Zafirovic SS, Radak DJ, Essack MM, et al. Regulation of nitric oxide production in hypothyroidism. Biomed Pharmacother. 2020;124:109881. https://doi.org/10.1016/j.biopha.2020.109881.

Gonzalez FM, Shiva S, Vincent PS, Ringwood LA, Hsu LY, Hon YY, et al. Nitrite anion provides potent cytoprotective and antiapoptotic effects as adjunctive therapy to reperfusion for acute myocardial infarction. Circulation. 2008; 117(23):2986–94. https://doi.org/10.1161/circulationaha.107.748814.

Gorbach SL. Bismuth therapy in gastrointestinal diseases. Gastroenterology. 1990; 99(3):863–75. https://doi.org/10.1016/0016-5085(90)90983-8.

Green LC, Ruiz de Luzuriaga K, Wagner DA, Rand W, Istfan N, Young VR, et al. Nitrate biosynthesis in man. Proc Natl Acad Sci U S A. 1981. https://doi.org/10.1073/pnas.78.12.7764.

Hernández A, Schiffer TA, Ivarsson N, Cheng AJ, Bruton JD, Lundberg JO, et al. Dietary nitrate increases tetanic [Ca2+]i and contractile force in mouse fast-twitch muscle. J Physiol. 2012;590(15):3575–83. https://doi.org/10.1113/jphysiol.2012.232777.

Hord NG, Tang Y, Bryan NS. Food sources of nitrates and nitrites: the physiologic context for potential health benefits. Am J Clin Nutr. 2009;90(1):1–10. https://doi.org/10.3945/ajcn.2008.27131.

Hu L, Jin L, Xia D, Zhang Q, Ma L, Zheng H, et al. Nitrate ameliorates dextran sodium sulfate-induced colitis by regulating the homeostasis of the intestinal microbiota. Free Radic Biol Med. 2020;152:609–21. https://doi.org/10.1016/j.freeradbiomed.2019.12.002.

IARC monographs on the evaluation of carcinogenic risks to humans. Ingested nitrate and nitrite, and cyanobacterial peptide toxins. IARC Monogr Eval Carcinog Risks Hum. 2010;94(v-vii):1–412.

Iwata M, Inoue T, Asai Y, Hori K, Fujiwara M, Matsuo S, et al. The protective role of localized nitric oxide production during inflammation may be mediated by the heme oxygenase-1/carbon monoxide pathway. Biochem Biophys Rep. 2020;23:100790. https://doi.org/10.1016/j.bbrep.2020.100790.

Jackson JK, Patterson AJ, MacDonald-Wicks LK, Oldmeadow C, McEvoy MA. The role of inorganic nitrate and nitrite in cardiovascular disease risk factors: a systematic review and meta-analysis of human evidence. Nutr Rev. 2018; 76(5):348–71. https://doi.org/10.1093/nutrit/nuy005.

Jädert C, Petersson J, Massena S, Ahl D, Grapensparr L, Holm L, et al. Decreased leukocyte recruitment by inorganic nitrate and nitrite in microvascular inflammation and NSAID-induced intestinal injury. Free Radic Biol Med. 2012; 52(3):683–92. https://doi.org/10.1016/j.freeradbiomed.2011.11.018.

Jädert C, Phillipson M, Holm L, Lundberg JO, Borniquel S. Preventive and therapeutic effects of nitrite supplementation in experimental inflammatory

bowel disease. Redox Biol. 2014;2:73–81. https://doi.org/10.1016/j.redox.2013.12.012.
Jin L, Qin L, Xia D, Liu X, Fan Z, Zhang C, et al. Active secretion and protective effect of salivary nitrate against stress in human volunteers and rats. Free Radic Biol Med. 2013;57:61–7. https://doi.org/10.1016/j.freeradbiomed.2012.12.015.
Jin L, Zhang M, Xu J, Xia D, Zhang C, Wang J, et al. Music stimuli lead to increased levels of nitrite in unstimulated mixed saliva. Sci China Life Sci. 2018;61(9):1099–106. https://doi.org/10.1007/s11427-018-9309-3.
Jones AM, Thompson C, Wylie LJ, Vanhatalo A. Dietary nitrate and physical performance. Annu Rev Nutr. 2018;38(1):303–28. https://doi.org/10.1146/annurev-nutr-082117-051622.
Kapil V, Khambata RS, Jones DA, Rathod K, Primus C, Massimo G, et al. The noncanonical pathway for in vivo nitric oxide generation: the nitrate-nitrite-nitric oxide pathway. Pharmacol Rev. 2020;72(3):692–766. https://doi.org/10.1124/pr.120.019240.
Kapil V, Khambata RS, Robertson A, Caulfield MJ, Ahluwalia A. Dietary nitrate provides sustained blood pressure lowering in hypertensive patients: a randomized, phase 2, double-blind, placebo-controlled study. Hypertension. 2015;65(2):320–7. https://doi.org/10.1161/hypertensionaha.114.04675.
Kapil V, Milsom AB, Okorie M, Maleki-Toyserkani S, Akram F, Rehman F, et al. Inorganic nitrate supplementation lowers blood pressure in humans: role for nitrite-derived NO. Hypertension. 2010;56(2):274–81. https://doi.org/10.1161/hypertensionaha.110.153536.
Khalifi S, Rahimipour A, Jeddi S, Ghanbari M, Kazerouni F, Ghasemi A. Dietary nitrate improves glucose tolerance and lipid profile in an animal model of hyperglycemia. Nitric Oxide. 2015;44:24–30. https://doi.org/10.1016/j.niox.2014.11.011.
Kina-Tanada M, Sakanashi M, Arasaki A, Tsutsui M. Long-term dietary nitrite and nitrate deficiency causes metabolic syndrome, endothelial dysfunction, and cardiovascular death in mice. Nihon Yakurigaku Zasshi. 2018. https://doi.org/10.1254/fpj.151.148.
Laakso J, Mervaala E, Himberg JJ, Teräväinen TL, Karppanen H, Vapaatalo H, et al. Increased kidney xanthine oxidoreductase activity in salt-induced experimental hypertension. Hypertension. 1998;32(5):902–6. https://doi.org/10.1161/01.hyp.32.5.902.
Larsen FJ, Weitzberg E, Lundberg JO, Ekblom B. Effects of dietary nitrate on oxygen cost during exercise. Acta Physiol (Oxford). 2007;191(1):59–66. https://doi.org/10.1111/j.1748-1716.2007.01713.x.
Li H, Samouilov A, Liu X, Zweier JL. Characterization of the effects of oxygen on xanthine oxidase-mediated nitric oxide formation. J Biol Chem 2004; https://doi.org/10.1074/jbc. M314336200.
Li S, An W, Wang B, Li J, Qu Y, Zhang H, et al. Inorganic nitrate alleviates irradiation-induced salivary gland damage by inhibiting pyroptosis. Free Radic Biol Med. 2021b;175:130–40. https://doi.org/10.1016/j.freeradbiomed.2021.08.227.
Li S, Jin H, Sun G, Zhang C, Wang J, Xu H, et al. Dietary inorganic nitrate protects hepatic ischemia-reperfusion injury through NRF2-mediated antioxidative stress. Front Pharmacol. 2021a;12. https://doi.org/10.3389/fphar.2021.634115.
Li T, Lu X, Sun Y, Yang X. Effects of spinach nitrate on insulin resistance, endothelial dysfunction markers and inflammation in mice with high-fat and high-fructose consumption. Food Nutr Res. 2016;60(1):32010. https://doi.org/10.3402/fnr.v60.32010.
Linder N, Rapola J, Raivio KO. Cellular expression of xanthine oxidoreductase protein in normal human tissues. Lab Investig. 1999;79(8):967–74.
Lundberg JO. Nitrate transport in salivary glands with implications for NO homeostasis. Proc Natl Acad Sci U S A. 2012;109(33):13144–5. https://doi.org/10.1073/pnas.1210412109.
Lundberg JO, Carlström M, Weitzberg E. Metabolic effects of dietary nitrate in health and disease. Cell Metab. 2018;28(1):9–22. https://doi.org/10.1016/j.cmet.2018.06.007.
Lundberg JO, Gladwin MT, Weitzberg E. Strategies to increase nitric oxide signalling in cardiovascular disease. Nat Rev Drug Discov. 2015;14(9):623–41. https://doi.org/10.1038/nrd4623.
Lundberg JO, Govoni M. Inorganic nitrate is a possible source for systemic generation of nitric oxide. Free Radic Biol Med. 2004;37(3):395–400. https://doi.org/10.1016/j.freeradbiomed.2004.04.027.
Lundberg JO, Weitzberg E, Gladwin MT. The nitrate-nitrite-nitric oxide pathway in physiology and therapeutics. Nat Rev Drug Discov. 2008;7(2):156–67. https://doi.org/10.1038/nrd2466.
Lundberg JO, Weitzberg E, Lundberg JM, Alving K. Intragastric nitric oxide production in humans: measurements in expelled air. Gut. 1994;35(11):1543–6. https://doi.org/10.1136/gut.35.11.1543.
Ma L, Hu L, Feng X, Wang S. Nitrate and nitrite in health and disease. Aging Dis. 2018;9(5):938–45. https://doi.org/10.14336/ad.2017.1207.
Ma L, Hu L, Jin L, Wang J, Li X, Wang W, et al. Rebalancing glucolipid metabolism and gut microbiome dysbiosis by nitrate-dependent alleviation of high-fat diet-induced obesity. BMJ Open Diabetes Res Care. 2020;8(1):e001255. https://doi.org/10.1136/bmjdrc-2020-001255.
McKnight GM, Smith LM, Drummond RS, Duncan CW, Golden M, Benjamin N. Chemical synthesis of nitric oxide in the stomach from dietary nitrate in humans. Gut. 1997;40(2):211–4. https://doi.org/10.1136/gut.40.2.211.
Millar TM, Stevens CR, Benjamin N, Eisenthal R, Harrison R, Blake DR. Xanthine oxidoreductase catalyses the reduction of nitrates and nitrite to nitric oxide under hypoxic conditions. FEBS Lett. 1998;427(2):225–8. https://doi.org/10.1016/s0014-5793(98)00430-x.
Milton-Laskibar I, Martínez JA, Portillo MP. Current knowledge on beetroot bioactive compounds: role of nitrate and betalains in health and disease. Foods. 2021;10(6). https://doi.org/10.3390/foods10061314.
Moncada S, Higgs A. The L-arginine-nitric oxide pathway. N Engl J Med. 1993;329(27):2002–12. https://doi.org/10.1056/nejm199312303292706.
Morris RC Jr, Pravenec M, Šilhavý J, DiCarlo SE, Kurtz TW. Small amounts of inorganic nitrate or beetroot provide substantial protection from salt-induced increases in blood pressure. Hypertension. 2019;73(5):1042–8. https://doi.org/10.1161/hypertensionaha.118.12234.
Münzel T, Daiber A, Mülsch A. Explaining the phenomenon of nitrate tolerance. Circ Res. 2005;97(7):618–28. https://doi.org/10.1161/01.RES.0000184694.03262.6d.
Ohtake K, Koga M, Uchida H, Sonoda K, Ito J, Uchida M, et al. Oral nitrite ameliorates dextran sulfate sodium-induced acute experimental colitis in mice. Nitric Oxide. 2010;23(1):65–73. https://doi.org/10.1016/j.niox.2010.04.004.
Pandey CB, Kumar U, Kaviraj M, Minick KJ, Mishra AK, Singh JS. DNRA: a short-circuit in biological N-cycling to conserve nitrogen in terrestrial ecosystems. Sci Total Environ. 2020;738:139710. https://doi.org/10.1016/j.scitotenv.2020.139710.
Park JW, Piknova B, Huang PL, Noguchi CT, Schechter AN. Effect of blood nitrite and nitrate levels on murine platelet function. PLoS One. 2013;8(2). https://doi.org/10.1371/journal.pone.0055699.
Paulus WJ, Tschöpe C. A novel paradigm for heart failure with preserved ejection fraction: comorbidities drive myocardial dysfunction and remodeling through coronary microvascular endothelial inflammation. J Am Coll Cardiol. 2013;62(4):263–71. https://doi.org/10.1016/j.jacc.2013.02.092.
Peleli M, Hezel M, Zollbrecht C, Persson AE, Lundberg JO, Weitzberg E, et al. In adenosine A2B knockouts acute treatment with inorganic nitrate improves glucose disposal, oxidative stress, and AMPK signaling in the liver. Front Physiol. 2015;6. https://doi.org/10.3389/fphys.2015.00222.
Pellegrino D, Shiva S, Angelone T, Gladwin MT, Tota B. Nitrite exerts potent negative inotropy in the isolated heart via eNOS-independent nitric oxide generation and cGMP-PKG pathway activation. Biochim Biophys Acta. 2009;1787(7):818–27. https://doi.org/10.1016/j.bbabio.2009.02.007.
Qin L, Liu X, Sun Q, Fan Z, Xia D, Ding G, et al. Sialin (SLC17A5) functions as a nitrate transporter in the plasma membrane. Proc Natl Acad Sci U S A. 2012;109(33):13434–9. https://doi.org/10.1073/pnas.1116633109.
Radomski MW, Palmer RM, Moncada S. Endogenous nitric oxide inhibits human platelet adhesion to vascular endothelium. Lancet. 1987;330(8567):1057–8. https://doi.org/10.1016/s0140-6736(87)91481-4.
Radomski MW, Palmer RM, Moncada S. An L-arginine/nitric oxide pathway present in human platelets regulates aggregation. Proc Natl Acad Sci U S A. 1990;87(13):5193–7. https://doi.org/10.1073/pnas.87.13.5193.
Rammos C, Hendgen-Cotta UB, Sobierajski J, Bernard A, Kelm M, Rassaf T. Dietary nitrate reverses vascular dysfunction in older adults with moderately increased cardiovascular risk. J Am Coll Cardiol. 2014;63(15):1584–5. https://doi.org/10.1016/j.jacc.2013.08.691.
Raubenheimer K, Bondonno C, Blekkenhorst L, Wagner KH, Peake JM, Neubauer O. Effects of dietary nitrate on inflammation and immune function, and implications for cardiovascular health. Nutr Rev. 2019;77(8):584–99. https://doi.org/10.1093/nutrit/nuz025.
Roberts LD, Ashmore T, Kotwica AO, Murfitt SA, Fernandez BO, Feelisch M, et al. Inorganic nitrate promotes the browning of white adipose tissue through

the nitrate-nitrite-nitric oxide pathway. Diabetes. 2015;64(2):471–84. https://doi.org/10.2337/db14-0496.
Sabbah DA, Hajjo R, Sweidan K. Review on epidermal growth factor receptor (EGFR) structure, signaling pathways, interactions, and recent updates of EGFR inhibitors. Curr Top Med Chem. 2020;20(10):815–34. https://doi.org/10.2174/1568026620666200303123102.
Samal AA, Honavar J, Brandon A, Bradley KM, Doran S, Liu Y, et al. Administration of nitrite after chlorine gas exposure prevents lung injury: effect of administration modality. Free Radic Biol Med. 2012;53(7):1431–9. https://doi.org/10.1016/j.freeradbiomed.2012.08.007.
Sampaio-Maia B, Caldas IM, Pereira ML, Pérez-Mongiovi D, Araujo R. The oral microbiome in health and its implication in oral and systemic diseases. Adv Appl Microbiol. 2016. https://doi.org/10.1016/bs.aambs.2016.08.002.
Sengupta S, Sellers LA, Gherardi E, Sasisekharan R, Fan TP. Nitric oxide modulates hepatocyte growth factor/scatter factor-induced angiogenesis. Angiogenesis. 2004;7(4):285–94. https://doi.org/10.1007/s10456-004-5238-3.
Sessa WC. Molecular control of blood flow and angiogenesis: role of nitric oxide. J Thromb Haemost. 2009;7:35–7. https://doi.org/10.1111/j.1538-7836.2009.03424.x.
Shiva S, Wang X, Ringwood LA, Xu X, Yuditskaya S, Annavajjhala V, et al. Ceruloplasmin is a NO oxidase and nitrite synthase that determines endocrine NO homeostasis. Nat Chem Biol. 2006;2(9):486–93. https://doi.org/10.1038/nchembio813.
Sinha R, Zhao N, Goedert JJ, Byrd DA, Wan Y, Hua X, et al. Effects of processed meat and drinking water nitrate on oral and fecal microbial populations in a controlled feeding study. Environ Res. 2021;197:111084. https://doi.org/10.1016/j.envres.2021.111084.
Sobko T, Marcus C, Govoni M, Kamiya S. Dietary nitrate in Japanese traditional foods lowers diastolic blood pressure in healthy volunteers. Nitric Oxide. 2010;22(2):136–40. https://doi.org/10.1016/j.niox.2009.10.007.
Sparacino-Watkins C, Stolz JF, Basu P. Nitrate and periplasmic nitrate reductases. Chem Soc Rev. 2014;43(2):676–706. https://doi.org/10.1039/c3cs60249d.
Stefanadis C, Antoniou CK, Tsiachris D, Pietri P. Coronary atherosclerotic vulnerable plaque: current perspectives. J Am Heart Assoc. 2017;6(3). https://doi.org/10.1161/jaha.117.005543.
Suhr F, Gehlert S, Grau M, Bloch W. Skeletal muscle function during exercise-fine-tuning of diverse subsystems by nitric oxide. Int J Mol Sci. 2013;14(4):7109–39. https://doi.org/10.3390/ijms14047109.
Sultan S, El-Mowafy M, Elgaml A, Ahmed TAE, Hassan H, Mottawea W. Metabolic influences of gut microbiota dysbiosis on inflammatory bowel disease. Front Physiol. 2021;12. https://doi.org/10.3389/fphys.2021.715506.
Sun Y, Liu WZ, Liu T, Feng X, Yang N, Zhou HF. Signaling pathway of MAPK/ERK in cell proliferation, differentiation, migration, senescence and apoptosis. J Recept Signal Transduct Res. 2015;35(6):600–4. https://doi.org/10.3109/10799893.2015.1030412.
Tang W, Yao X, Xia F, Yang M, Chen Z, Zhou B, et al. Modulation of the gut microbiota in rats by hugan qingzhi tablets during the treatment of high-fat-diet-induced nonalcoholic fatty liver disease. Oxidative Med Cell Longev. 2018;2018:1–14. https://doi.org/10.1155/2018/7261619.
Tannenbaum SR, Weisman M, Fett D. The effect of nitrate intake on nitrite formation in human saliva. Food Cosmet Toxicol. 1976;14(6):549–52. https://doi.org/10.1016/s0015-6264(76)80006-5.
Tarkin J, Kaski J. Vasodilator therapy: nitrates and Nicorandil. Cardiovasc Drugs Ther. 2016;30(4):367–78. https://doi.org/10.1007/s10557-016-6668-z.
Tominaga H, Kodama S, Matsuda N, Suzuki K, Watanabe M. Involvement of reactive oxygen species (ROS) in the induction of genetic instability by radiation. J Radiat Res. 2004;45(2):181–8. https://doi.org/10.1269/jrr.45.181.
Toprakçi M, Ozmen D, Mutaf I, Turgan N, Parildar Z, Habif S, et al. Age-associated changes in nitric oxide metabolites nitrite and nitrate. Int J Clin Lab Res. 2000;30(2):83–5. https://doi.org/10.1007/bf02874163.
van Heerebeek L, Hamdani N, Falcão-Pires I, Leite-Moreira AF, Begieneman MP, Bronzwaer JG, et al. Low myocardial protein kinase G activity in heart failure with preserved ejection fraction. Circulation. 2012;126(7):830–9. https://doi.org/10.1161/circulationaha.111.076075.
van Velzen AG, Sips AJ, Schothorst RC, Lambers AC, Meulenbelt J. The oral bioavailability of nitrate from nitrate-rich vegetables in humans. Toxicol Lett. 2008;181(3):177–81. https://doi.org/10.1016/j.toxlet.2008.07.019.
Vanhatalo A, Blackwell JR, L'Heureux JE, Williams DW, Smith A, van der Giezen M, et al. Nitrate-responsive oral microbiome modulates nitric oxide homeostasis and blood pressure in humans. Free Radic Biol Med. 2018;124:21–30. https://doi.org/10.1016/j.freeradbiomed.2018.05.078.
Velmurugan S, Gan JM, Rathod KS, Khambata RS, Ghosh SM, Hartley A, et al. Dietary nitrate improves vascular function in patients with hypercholesterolemia: a randomized, double-blind, placebo-controlled study. Am J Clin Nutr. 2016;103(1):25–38. https://doi.org/10.3945/ajcn.115.116244.
Wang H, Hu L, Li L, Wu X, Fan Z, Zhang C, et al. Inorganic nitrate alleviates the senescence-related decline in liver function. Sci China Life Sci. 2018;61(1):24–34. https://doi.org/10.1007/s11427-017-9207-x.
Wang W, Hu L, Chang S, Ma L, Li X, Yang Z, et al. Total body irradiation-induced colon damage is prevented by nitrate-mediated suppression of oxidative stress and homeostasis of the gut microbiome. Nitric Oxide. 2020;102:1–11. https://doi.org/10.1016/j.niox.2020.05.002.
Webb AJ, Patel N, Loukogeorgakis S, Okorie M, Aboud Z, Misra S, et al. Acute blood pressure lowering, vasoprotective, and antiplatelet properties of dietary nitrate via bioconversion to nitrite. Hypertension. 2008;51(3):784–90. https://doi.org/10.1161/hypertensionaha.107.103523.
Weitzberg E, Lundberg JO. Novel aspects of dietary nitrate and human health. Annu Rev Nutr. 2013;33(1):129–59. https://doi.org/10.1146/annurev-nutr-071812-161159.
Wilson DF. Factors affecting the rate and energetics of mitochondrial oxidative phosphorylation. Med Sci Sports Exerc. 1994;26(1):37–43. https://doi.org/10.1249/00005768-199405001-00213.
Xia D, Deng D, Wang S. Alterations of nitrate and nitrite content in saliva, serum, and urine in patients with salivary dysfunction. J Oral Pathol Med. 2003;32(2):95–9. https://doi.org/10.1034/j.1600-0714.2003.00109.x.
Xu Y, Pang B, Hu L, Feng X, Hu L, Wang J, et al. Dietary nitrate protects submandibular gland from hyposalivation in ovariectomized rats via suppressing cell apoptosis. Biochem Biophys Res Commun. 2018;497(1):272–8. https://doi.org/10.1016/j.bbrc.2018.02.068.
Yang T, Zhang XM, Tarnawski L, Peleli M, Zhuge Z, Terrando N, et al. Dietary nitrate attenuates renal ischemia-reperfusion injuries by modulation of immune responses and reduction of oxidative stress. Redox Biol. 2017;13:320–30. https://doi.org/10.1016/j.redox.2017.06.002.
Zamani P, Rawat D, Shiva-Kumar P, Geraci S, Bhuva R, Konda P, et al. Effect of inorganic nitrate on exercise capacity in heart failure with preserved ejection fraction. Circulation. 2015;131(4):371–80. https://doi.org/10.1161/circulationaha.114.012957.
Zhang X, Tang N, Hadden TJ, Rishi AK. Akt, FoxO and regulation of apoptosis. Biochim Biophys Acta. 2011;1813(11):1978–86. https://doi.org/10.1016/j.bbamcr.2011.03.010.
Zhu SG, Kukreja RC, Das A, Chen Q, Lesnefsky EJ, Xi L. Dietary nitrate supplementation protects against doxorubicin-induced cardiomyopathy by improving mitochondrial function. J Am Coll Cardiol. 2011;57(21):2181–9. https://doi.org/10.1016/j.jacc.2011.01.024.

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